

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Division of Patent Application Serial No. 08/696,988 of

YAMASAKI et al

Atty. Ref.: 249-159

Serial No. to be assigned

Group:

Filed: January 30, 2001

Examiner:

For: PLATELET PRODUCTION PROMOTING AGENT

* * * * *

January 29, 2001

Assistant Commissioner for Patents
Washington, DC 20231

Sir:

PRELIMINARY AMENDMENT

In order to place the above-identified application in better condition for examination, please amend the application as follows:

IN THE CLAIMS

Cancel claims 1 through 14 and add the following new claims:

15. (New) A modified polypeptide having human granulocyte colony stimulating factor activity (hG-CSF),
- (a) wherein at least one group of the carboxyl, mercapto or guanidino group in the molecule of the polypeptide is chemically modified with a polyalkylene glycol derivative; or
- (b) wherein the polypeptide has the amino acid sequence of SEQ ID NO:1, wherein at least one group of the amino, carboxyl, mercapto or guanidino group in the molecule of the polypeptide is chemically modified with a polyalkylene glycol derivative.

16. (New) The modified polypeptide according to claim 15,

- (a) wherein at least one amino acid of the 1st to 6th and 17th amino acids from the N terminus side in the amino acid sequence of SEQ ID NO:1 is replaced by another amino acid; or
- (b) wherein at least one of the 1st to 11th amino acids from the N terminus side in the amino acid sequence of SEQ ID NO:1 is deleted and the 17th amino acid thereof is optionally replaced by Ser.

17. (New) The modified polypeptide according to claim 15, wherein the amino acid sequence of the polypeptide has, from the N terminus,

- (a) Glu in the third position, Lys in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (b) Val in the first position, Ile in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (c) Cys in the first position, Ile in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (d) Tyr in the first position, Ile in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (e) Arg in the first position, Thr in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (f) Thr in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (g) Asn in the first position, Glu in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (h) Ile in the first position, Thr in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,

- (i) Ser in the first position, Thr in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (j) Arg in the fourth position and Ser in the seventeenth position,
- (k) Ala in the first position, Thr in the third position, Tyr in the fourth position, Arg in the fifth position and Ser in the seventeenth position, or
- (l) Ser in the seventeenth position.

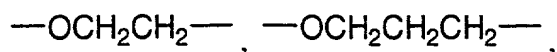
18. The modified polypeptide according to claim 15, wherein the amino acid sequence of the polypeptide has Ala in the first position, Thr in the third position, Tyr in the fourth position, Arg in the fifth position and Ser in the seventeenth position from the N terminus.

19. The modified polypeptide according to claim 15, wherein the polyalkylene glycol derivative is selected from the consisting of a polyethylene glycol derivative, polypropylene glycol derivative, and a derivative of polyethylene glycol-polypropylene glycol copolymer.

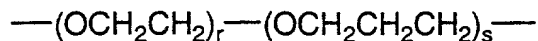
20. The modified polypeptide according to claim 15, wherein the amino group is modified with a polyalkylene glycol derivative having the formula (I):



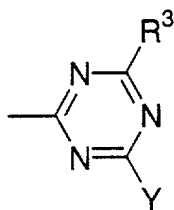
wherein R^1 represents an alkyl or alkanoyl group; M represents the formula:



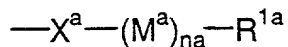
or



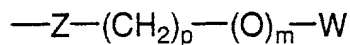
wherein r and s have any variable positive integral values, which are the same or different; n has any variable positive integral values; X represents a single bond, O, NH, or S; and R^2 represents the formula:



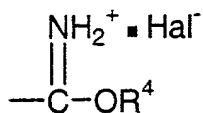
wherein R³ represents OH, halogen, or the formula:



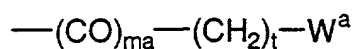
wherein X^a, M^a, R^{1a} and n_a are identical to said X, M, R¹ and n, respectively, and Y represents halogen or the formula:



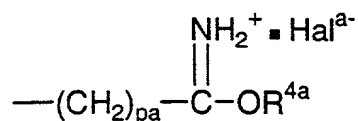
wherein Z represents O, S, or NH; W represents a carboxyl group, an active derivative thereof, or the formula:



wherein R⁴ represents an alkyl group; and Hal represents halogen, and p has an integral value of 1 to 6; and m has a value of 0 or 1,



wherein W^a and m_a are identical to said W and m, respectively; and t has an integral value of from 0 to 6, or



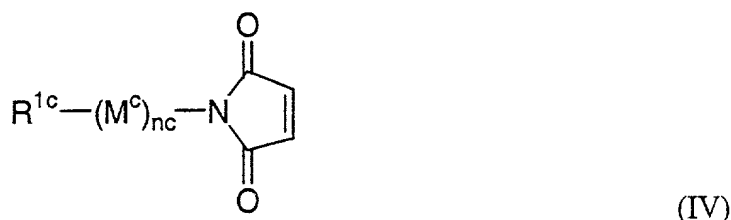
wherein Hal^a, p_a and R^{4a} are identical to said Hal, p and R⁴, respectively.

21. (New) The modified polypeptide according to claim 15, wherein the carboxyl group is modified with a polyalkylene glycol derivative having the formula (III):



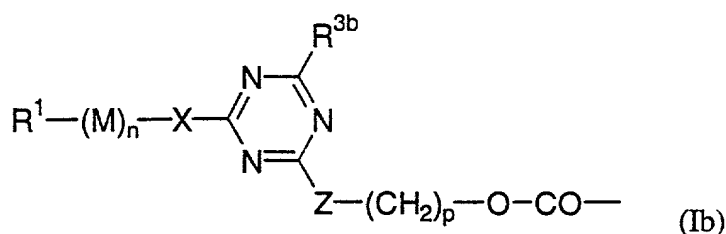
wherein M^b , R^{1b} and nb are identical to said M , R^1 and n , respectively.

22. (New) The modified polypeptide according to claim 15, wherein the mercapto group is modified with a polyalkylene glycol derivative having the formula (IV):

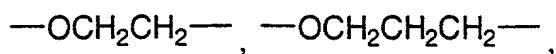


wherein M^c , R^{1c} , and nc are identical to said M , R^1 , and n , respectively.

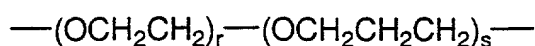
23. (New) A modified polypeptide having human granulocyte colony stimulating factor activity (hG-CSF), wherein at least one group amino group in the formula is substituted with a group of the formula (Ib):



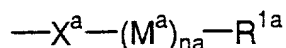
wherein R^1 represents an alkyl or alkanoyl group; M represents the formula:



or



wherein r and s have any variable positive integral values, which are the same or different, n has any variable positive integral value; X represents a single bond, O, NH, or S; R^{3b} represents OH, halogen, or the formula:



wherein X^a, M^a, R^{1a} and n_a are identical to said X, M, R¹ and n, respectively; Z represents O, S, or NH; and p has an integral value of from 1 to 6.

24. A method for treating a patient with decreased platelet counts comprising administering an effective amount of:

- (a) a modified polypeptide having human granulocyte colony stimulating factor activity (hG-CSF), wherein at least one group of the amino, carboxyl, mercapto or guanidino group in the molecule of the polypeptide is chemically modified with a polyalkylene glycol derivative or a styrene-maleic acid copolymer;
- (b) a modified polypeptide having human granulocyte colony stimulating factor activity (hG-CSF) having the amino acid sequence of SEQ ID NO:1, wherein at least one group of the amino, carboxyl, mercapto or guanidino group in the molecule of the polypeptide is chemically modified with a polyalkylene glycol derivative or a styrene-maleic acid copolymer; or
- (c) the modified polypeptide of claim 23.

25. (New) The method according to claim 24,

- (a) wherein at least one amino acid of the 1st to 6th and 17th amino acids from the N terminus side in the amino acid sequence of SEQ ID NO:1 is replaced by another amino acid; or
- (b) wherein at least one of the 1st to 11th amino acids from the N terminus side in the amino acid sequence of SEQ ID NO:1 is deleted and the 17th amino acid thereof is optionally replaced by Ser.

26. (New) The method according to claim 24, wherein the amino acid sequence of the polypeptide has, from the N terminus,

- (a) Glu in the third position, Lys in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (b) Val in the first position, Ile in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (c) Cys in the first position, Ile in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (d) Tyr in the first position, Ile in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (e) Arg in the first position, Thr in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (f) Thr in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (g) Asn in the first position, Glu in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (h) Ile in the first position, Thr in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (i) Ser in the first position, Thr in the third position, Arg in the fourth position, Ser in the fifth position and Ser in the seventeenth position,
- (j) Arg in the fourth position and Ser in the seventeenth position,
- (k) Ala in the first position, Thr in the third position, Tyr in the fourth position, Arg in the fifth position and Ser in the seventeenth position, or
- (l) Ser in the seventeenth position.

27. (New) The method according to claim 24, wherein the amino acid sequence of the polypeptide has Ala in the first position, Thr in the third position, Tyr in the fourth position, Arg in the fifth position and Ser in the seventeenth position from the N terminus.

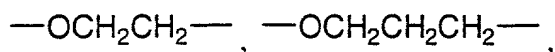
28. (New) The method according to claim 24, wherein the polyalkylene glycol derivative is selected from the group consisting of a polyethylene glycol derivative, a polypropylene glycol derivative, and a derivative of polyethylene glycol-polypropylene glycol copolymer.

29. (New) The method according to claim 24, wherein the amino group is modified with

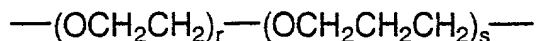
(a) a polyalkylene glycol derivative having the formula (I):



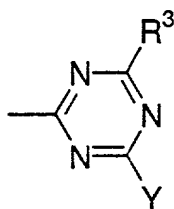
wherein R^1 represents an alkyl or alkanoyl group; M represents the formula:



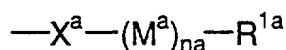
or



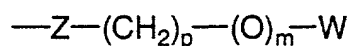
wherein r and s have any variable positive integral values, which are the same or different; n has any variable positive integral values; X represents a single bond, O, NH, or S; and R^2 represents the formula:



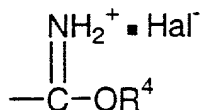
wherein R^3 represents OH, halogen, or the formula:



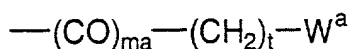
wherein X^a , M^a , R^{1a} and na are identical to said X, M, R^1 and n, respectively, and Y represents halogen or the formula:



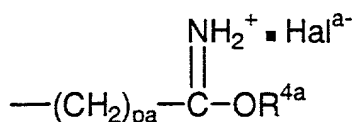
wherein Z represents O, S, or NH; W represents a carboxyl group, an active derivative thereof, or the formula:



wherein R^4 represents an alkyl group; and Hal represents halogen, and p has an integral value of 1 to 6; and m has a value of 0 or 1,

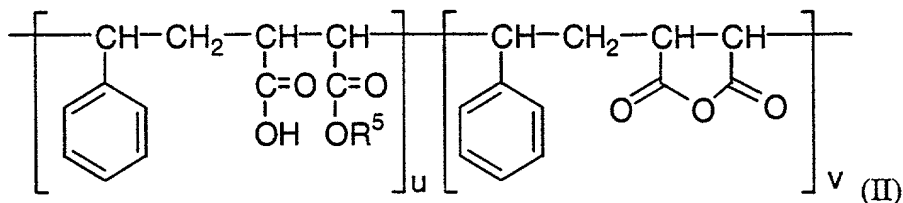


wherein W^a and ma are identical to said W and m, respectively; and t has an integral value of from 0 to 6, or



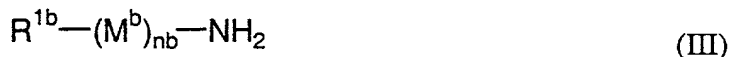
wherein Hal^a , pa and R^{4a} are identical to said Hal, p and R^4 , respectively, or

(b) a styrene-maleic acid copolymer having the formula (II):



wherein u and v have any variable positive integral values, which are the same or different; and R^5 represents a hydrogen atom, or an alkyl group.

30. (New) The method according to claim 24, wherein the carboxyl group is modified with a polyalkylene glycol derivative having the formula (III):

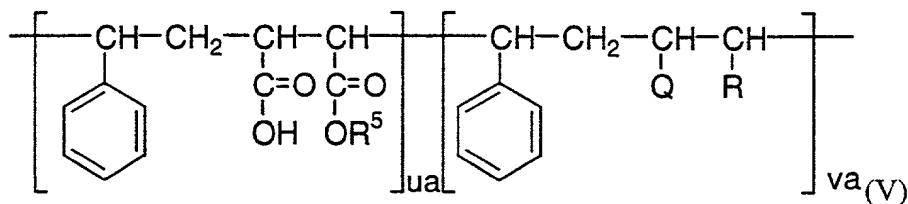


wherein M^b , R^{1b} and nb are identical to said M, R^1 and n, respectively.

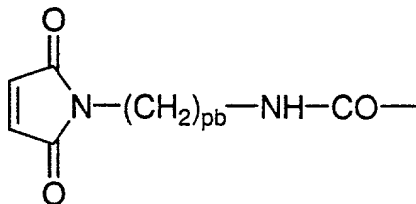
(a) a polyalkylene glycol derivative having the formula (IV):



(b) a styrene-maleic acid copolymer having the formula (V):

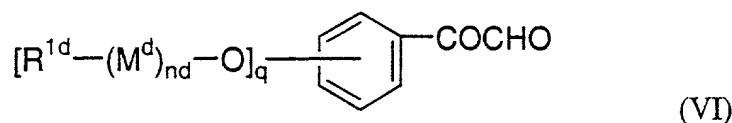


wherein R^{5a}, ua, and va are identical to said R⁵, U, and V, respectively, and one of Q and R represents a carboxyl group, and the other represents the formula:



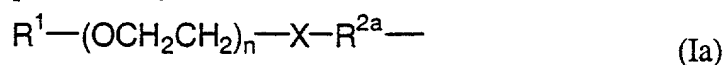
wherein pb is identical to said p.

32. (New) The method according to claim 24, wherein the guanidino group is modified with a polyalkylene glycol derivative having the formula (VI):

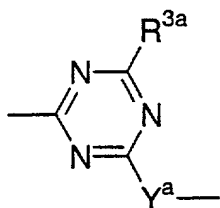


wherein q has a value of 1 or 2, and M^d , R^{1d} , and nd are identical to said M, R^1 , and n, respectively.

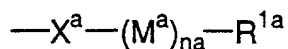
33. The method according to claim 24, wherein the amino group is modified by binding to a group represented by the formula (Ia):



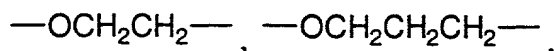
wherein R^1 represents an alkyl or alkanoyl group; n has any variable positive integral value; X represents a single bond, O, NH, or S; R^{2a} represents the formula:



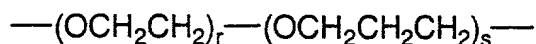
wherein R^{3a} represents OH, halogen, or the formula:



wherein X^a , R^{1a} and na are identical to said X, R^1 and n, respectively, M^a represents the formula:

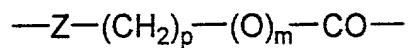


or



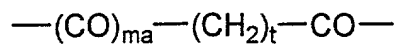
wherein r and s have any variable positive integral values, which are the same or different, and

Y^a represents a single bond, the formula:



wherein Z represents O, S, or NH; p has an integral value of from 1 to 6; and m has a value of 0

or 1, or the formula:



wherein ma is identical to said m; and t has an integral value of from 0 to 6.

REMARKS

This is a division of earlier application Serial No. 08/696,988 filed August 16, 1996 on which the issue fee has been paid. A new set of claims is presented for examination.

Claim 15 is directed to (a) the polypeptide in the originally filed claim 1 in which the chemically modifying agent is specified as a polyalkylene glycol derivative, and the groups to be modified are specified to be carboxyl, mercapto and guanidino groups. Claims 16-23 correspond to claims 18-20, 4-8 and 14, respectively, in which the chemically modifying agent is a polyalkylene glycol derivative.

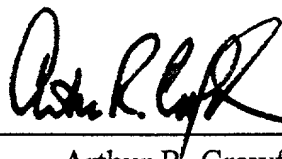
Also claim 24 is directed to a method of treatment using (a) the polypeptide in the originally filed claim 1, (b) the polypeptide in allowed claim 24, or (c) the polypeptide in new claim 23. Claims 25-33 correspond to claims 15-17 and claims 4-9 respectively.

Please examine these new claims taking into account the prior art of record in the parent application as listed in the attached Information Disclosure Statement.

Respectfully submitted,

NIXON & VANDERHYE P.C.

By: _____



Arthur R. Crawford
Reg. No. 25,327

ARC:pfc
1100 North Glebe Road, 8th Floor
Arlington, VA 22201-4714
Telephone: (703) 816-4000
Facsimile: (703) 816-4100